

Cell Biology: Function guides form of auditory sensory cells

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Summary

Mechanosensory bundles on auditory sensory cells are comprised of stereocilia that grow in rows of decreasing height. This pattern depends on specifying what will be the tallest row then assigning distinct molecular identities to the shorter rows. Mechanotransduction refines and maintains row identity, thus instructing the form of the bundle.

The bundles of stereocilia on sensory hair cells that detect sound in the inner ear are beautiful, probably because of their visual symmetry. Their function, however, depends to a greater degree on molecular asymmetry. It works like this: several stereocilia, which are actin-based protrusions, grow to form a bundle on top of auditory sensory cells. The stereocilia are arranged in tall, medium, and short rows, each of a particular height. Force-sensitive ion channels housed in the tips of the stereocilia in shorter rows are then connected to the sides of stereocilia in the next taller row by extracellular linkages. As the taller row is pushed backwards by sound energy, the channel opens and ions flood in, depolarizing the cell. This is mechanotransduction – converting physical movement into neuronal signals. Tiered rows of stereocilia are required for proper mechanotransduction, as the bundle shape influences its function. Two new studies by Tadenev *et al.* [1] and Krey *et al.* [2] provide exciting new insights into stereocilia bundle morphogenesis. Their data define key proteins needed to establish the molecular asymmetry between the stereocilia rows and reveal how mechanotransduction itself influences these proteins to refine bundle form.

The stereocilia bundle emerges during late embryonic mouse development from much thinner microvilli, which initially cover the entire surface of the hair cell. A subset of these protrusions elongate and widen to become stereocilia, while the remainder are resorbed [3]. By the time the mouse is born the tiered arrangement of stereocilia is evident, though still immature and unrefined. Within a few days of birth, the bundle starts to develop mechanotransduction currents [4], which seem to affect stereocilia development [2, 5]. In the chicken, it has long been understood that stereocilia have two periods of elongation separated by a widening phase [6]. A significant advance in the current work is to establish that mouse stereocilia development follows a similar progression and is coordinated with the onset of mechanotransduction. As ion flux increases, elongation pauses while stereocilia widen [2]. Several different proteins are essential for the bundle to develop to its normal dimensions, presumably contributing to and coordinating growth and mechanotransduction. In particular, a group of five proteins: Myo15a, Eps8, Whrn, GPSM2, and GNai3, appear essential to promote elongation of the tallest row and provide molecular asymmetry to the bundle [7-11].

A key insight of Tadenev *et al.* is that GPSM2, GNai3, and Whrn arrive after Myo15a and Eps8 to specify what will become the tallest row of the bundle from a group of similar microvilli. Multiple lines of evidence support this view. First, they performed an ambitious genetic analysis, which showed that double mutants lacking combinations of the five proteins were almost indistinguishable from any of the single mutants. This is strong evidence that these proteins are united in a common mission to facilitate growth of the tallest row (row 1). Each protein eventually localizes to the tip of row 1 stereocilia where they are positioned to direct

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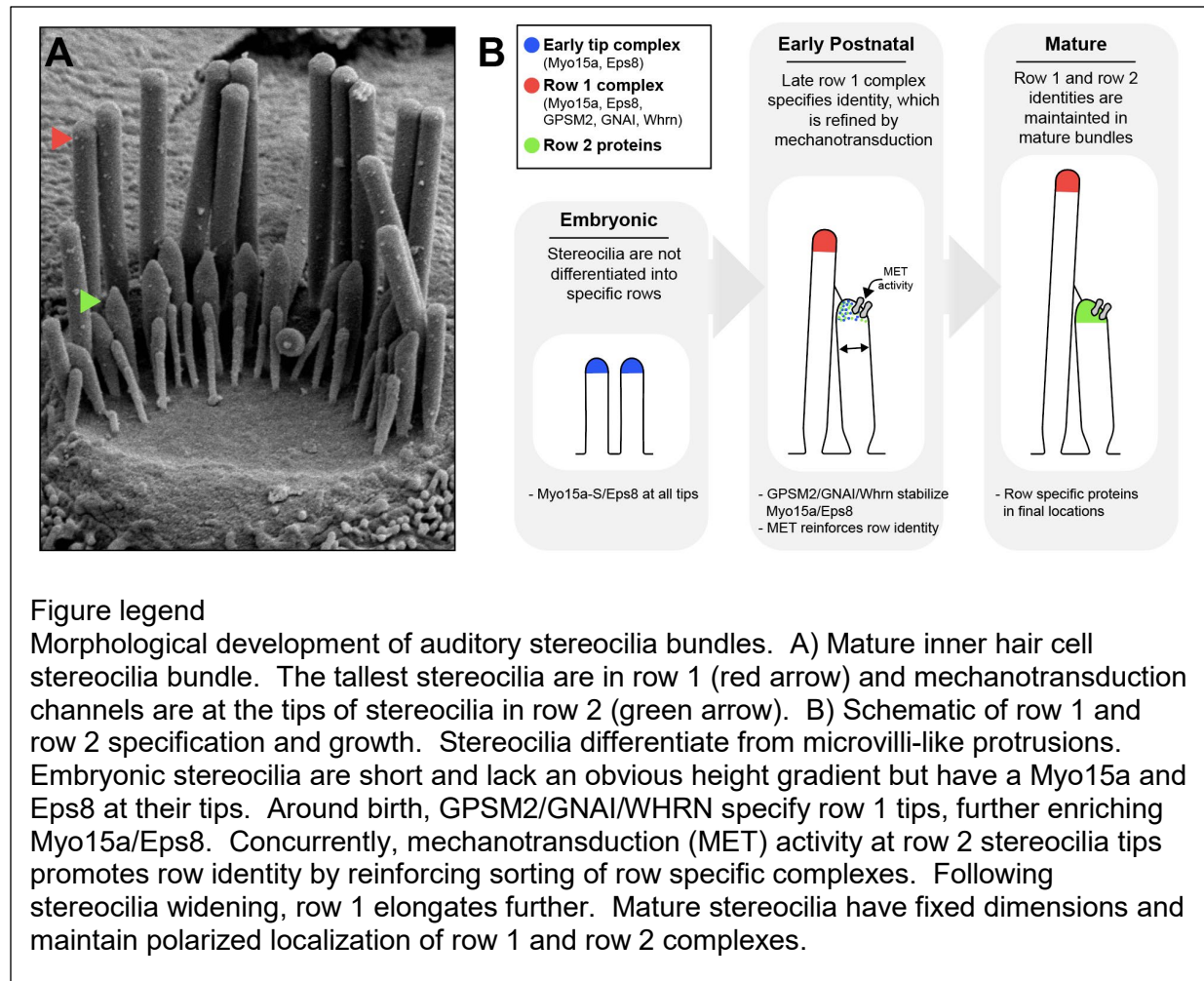
elongation, but the order of their arrival is highly informative. Myo15a and Eps8 are first on the scene. This pair is initially present at the tips of stereocilia in all rows, suggesting a general role in elongation. Next, GPSM2, GNai3 and Whrn more selectively target row 1 tips and together they reinforce Myo15a and Eps8 localization. As Myo15a and Eps8 are stabilized at row 1 tips their levels decrease in shorter stereocilia to produce a highly asymmetric localization pattern between rows.

When GPSM2 or GNai3 are deleted, row 1 identity is diluted as the remaining row 1 proteins now appear equally in all rows. Intriguingly, other proteins that are typically restricted to row 2 tips likewise find their way to row 1 tips [1]. Thus, failing to solidify row 1's distinct molecular identity results in all stereocilia settling into an intermediate, undifferentiated state, perhaps similar to the unrefined bundle observed at birth.

Krey *et al.* further advance understanding of stereocilia bundle development by considering the impact of mechanotransduction on morphogenesis [2]. At first glance, mutant mice lacking components of the mechanotransduction channel have normal looking bundles. However, closer inspection reveals notable differences including extra rows of stereocilia along with changes in both length and width [2, 5, 12, 13]. The work of Krey *et al.* provides a much more detailed understanding of the molecular events that follow loss of mechanotransduction [2]. Row 2 proteins can now be found in row 1. Shifting localization of row 2 proteins is not so surprising since they may be ordinarily enriched by mechanotransduction channels, themselves at row 2 but not row 1 tips [14]. Interestingly, row 1 proteins are also mislocalized without mechanotransduction. For example, Myo15a and Eps8, which typically define row 1 stereocilia are more abundantly localized to row 2 stereocilia in mechanotransduction mutants. Even more intriguing is that they found simply blocking the mechanotransduction channel causes a similar loss of row identity, suggesting that ion flux is a relevant upstream signal in this pathway. Thus, bundle development is a dynamic process with feedback from function guiding the ultimate form of the bundle through the sorting of proteins to either row 1 or row 2 stereocilia tips.

The overall molecular framework guiding stereocilia bundle morphogenesis is now clearer than ever. Nevertheless, many intriguing questions remain to challenge the field. For example, how does ion influx through channels at row 2 stereocilia tips change the localization of proteins at row 1 tips? Another missing piece is how row-specific proteins and mechanotransduction regulate the actin core of stereocilia to change their shape. The mechanism coupling these processes must use ion influx to regulate actin binding proteins and is at the heart of coordinating form with function. Hopefully future studies will address these knowledge gaps.

The processes that build the bundle may also take part in its maintenance. Certainly, mechanotransduction is well known to regulate the length of row 2 stereocilia. When the extracellular links that connect to the mechanotransduction channels are lost, or the mechanotransduction channel is simply blocked, then row 2 stereocilia rapidly shorten [15, 16]. In mature stereocilia, actin turnover is more evident in row 2 tips than elsewhere in stereocilia, perhaps reflecting repair following transient loss of mechanotransduction [17-19]. Better appreciating the connections between mechanotransduction function and stereocilia form promises to shed new light on both the development and maintenance of these remarkable and unique cell protrusions.



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